Figure legends for supplementary data

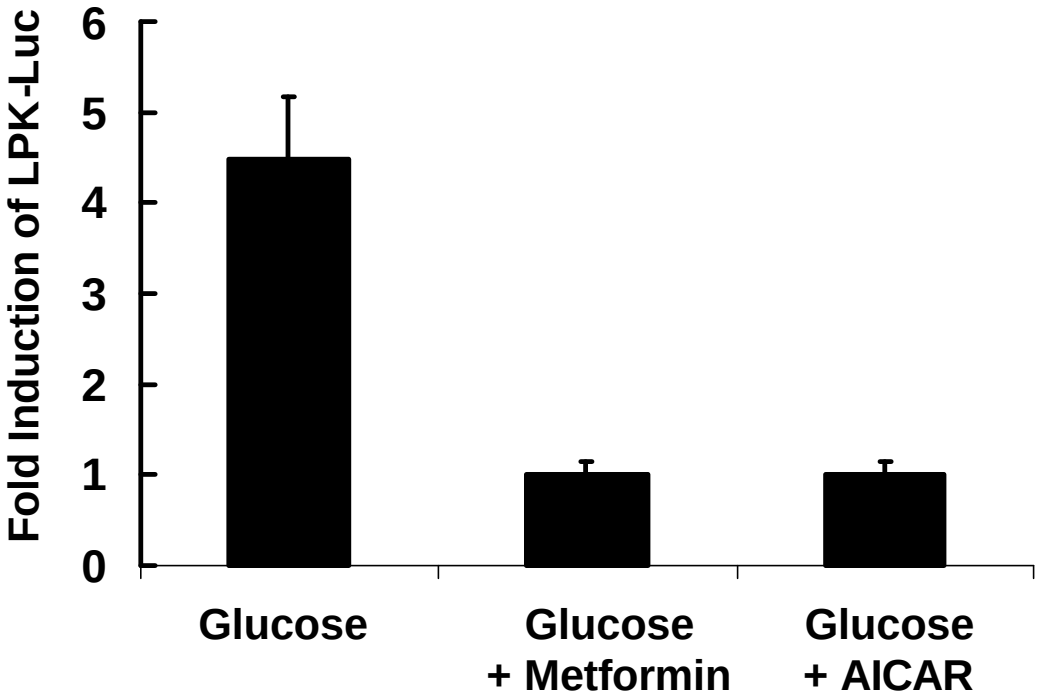
Figure 1. Effect of AMP-kinase (AMPK) activators on L-PK expression in primary rat hepatocytes. [**A**.] Primary hepatocytes were transfected with L-PK-Luc and phRG-Luc (Renilla Luc) while maintained in Williams E + lactate + insulin. Cells were switched to Williams E + glucose + insulin in the absence or presence of the AMPK activators, metformin (2 mM) or AICAR (2 mM). Twenty-four hours later cells were harvested for luciferase activity. Results are expressed as Fold Induction of L-PK-Luc activity, mean $\pm$ SD, N=6. [**B**]. Non-transfected primary hepatocytes were treated as above with glucose in the absence or presence of metformin (2 mM) and AICAR (2 mM) for the times indicated. Cells were harvested for RNA extraction and northern analysis for L-PK and 18S RNA. Results are representative of 2 separate experiments.

Figure 2. Effect of n-3 PUFA and WY14643 on AMPK and phospho-AMPK (pAMPK) in rat liver (A) and rat primary hepatocytes (B).
[**A**] Rat liver post-nuclear (cytosolic) and nuclear extracts were prepared from animals that were meal-fed high carbohydrate diets supplemented with olive oil, fish oil or olive oil + WY14643 for 7 days (Materials and Methods and Figures 2 and 3 of the text). Fasted (Fast) animals were meal fed the high carbohydrate + olive oil diet for 6 days and fasted overnight. Duplicate samples for each treatment were fractionated for immunoblotting with antibodies against phospho-AMPK (pAMPK), total AMPK, β-actin for post-nuclear (cytosolic) extracts and HNF-4α for nuclear extracts. Quantified results are represented as the ratio of pAMPK to total AMPK (pAMPK:AMPK), mean $\pm$ SD, N=4. Results are representative of 2 separate studies involving 7 separate animals/treatment group. *$p<0.05$ vs olive oil-fed, Students T-test.

[B] Primary hepatocytes from chow-fed (*ad lib*) rats were maintained in Williams E medium containing lactate + insulin (100 nM) or switched to Williams E medium containing glucose + insulin (100 nM) supplemented with 20:5,n-3 or WY14643 as described in Fig. 4. Figure 4 of the text illustrates the glucose-induced accumulation of ChREBP in hepatocyte nuclei. Cells were harvested at the times indicated for isolation of nuclear (shown) and post-nuclear (not shown) protein extracts. Proteins were fractionated for immunoblotting with antibodies against total AMPK and phospho-AMPK (pAMPK). Total and pAMPK levels were quantified from 3 separate studies. Results are expressed as the ratio of pAMPK to total AMPK (pAMPK:AMPK); mean $\pm$ SD, n=3. The results for the nuclear-extracts indicate that total AMPK and pAMPK levels were not significantly affected by 20:5,n-3 or WY14643 treatment. Similar results were obtained when post-nuclear (cytosolic) extracts were examined (not shown). Glucose (closed circles, solid line); Glucose + 20:5,n-3 (closed squares, solid line); Glucose + WY14643 (closed triangles + dashed line).

Figure 1 A & B

A. LPK-Luc Activity



B. Northern Blot

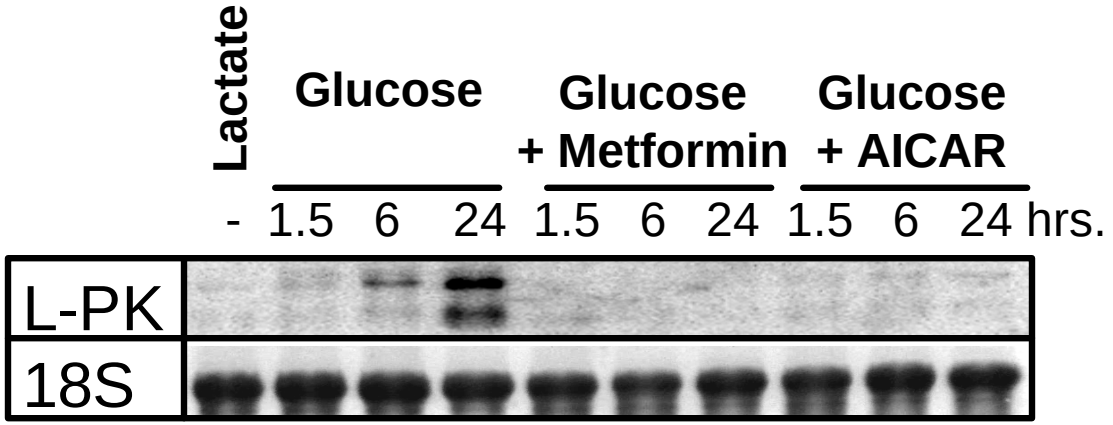
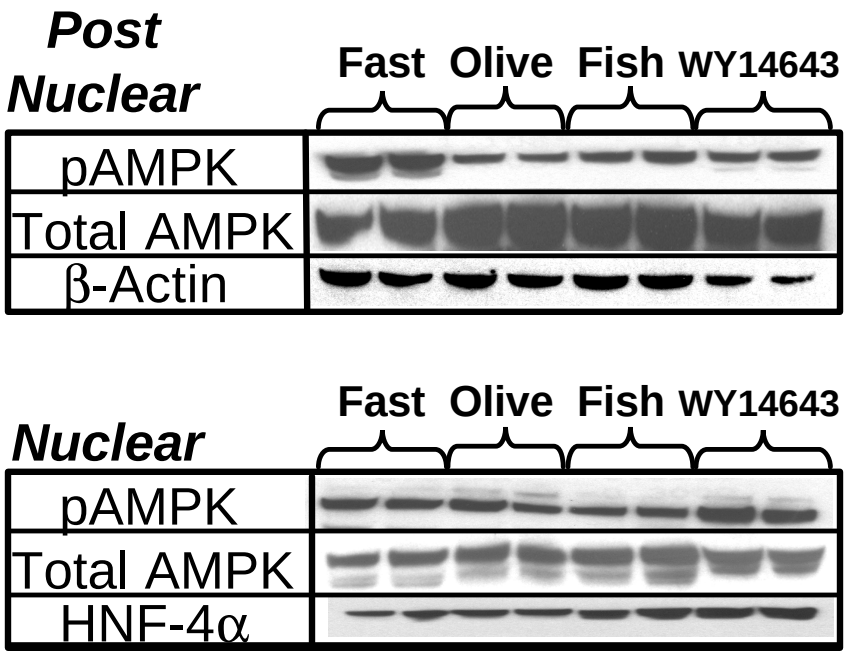


Figure 2A: AMPK in Rat Liver



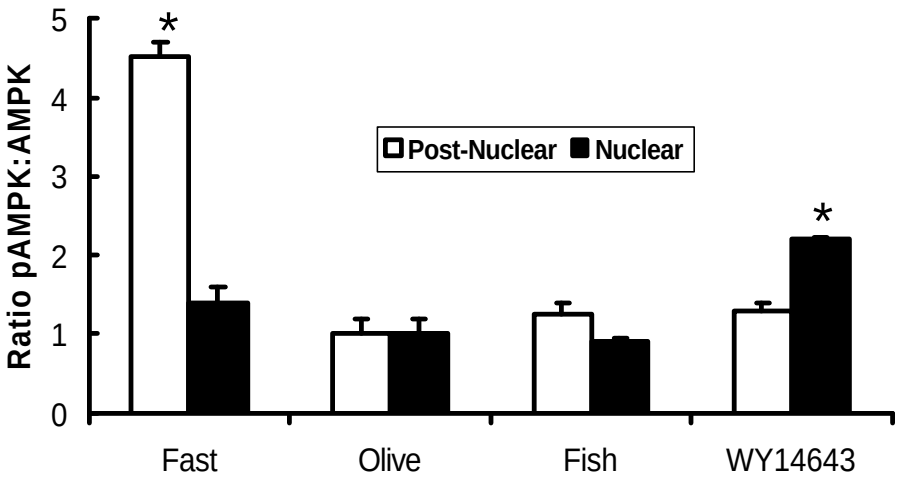


Figure 2B
AMPK in Primary Hepatocytes